

Claims

- Method for transforming plant cells by introducing a heterologous gene into said plant cells with a gene for tolerance to HPPD inhibitors as a selection marker, said
- 5 method comprising the steps of:
 - a) preparing and culturing competent plant cells capable of receiving the heterologous gene in a suitable medium,
 - b) transforming the competent cells with the heterologous gene and the selection marker,
 - 10 c) growing and selecting the transformed cells comprising the heterologous gene in a suitable medium,

characterized in that a step for bleaching the competent plant cells is carried out before the transformation step (b), by introducing a suitable amount of HPPD inhibitor into the suitable culture medium of the competent plant cells.
 - 15 2. Method for preparing transgenic plants comprising a heterologous gene integrated into their genome, comprising a method for transforming plant cells according to Claim 1, characterized in that it consists in carrying out the following steps of:
 - d) regenerating plants from the transformed cells selected in one or more suitable media and, where appropriate,
 - 20 e) producing and recovering the seeds of the fertile transformed plants.
 3. Method according to Claim 2, characterized in that the transgenic plants produced using the method according to the invention are fertile transgenic plants.
 - Method according to one of Claims 1 to 3, characterized in that the plant cells are chosen from the cells of dicotyledonous plants, in particular tobacco, rapeseed,
 - 25 sugar beet, potatoes, cotton and soya bean.
 5. Method according to Claim 4, characterized in that the plant cells are soya bean cells.
 - Method according to one of Claims 1 to 5, characterized in that the competent plant cells are chosen from embryogenic calluses, cell cultures on a solid
 - 30 support or in suspension, or embryogenic tissues.
 7. Method according to Claim 6, characterized in that the competent plant cells are proliferating embryogenic tissues.
 8. Method according to Claim 7, characterized in that the proliferating embryogenic tissues are maintained in a semi-solid medium.

9. Method according to Claim 8, characterized in that the semi-solid medium is an FNL medium.

10. Method according to one of Claims 1 to 10, characterized in that the HPPD inhibitor is chosen from isoxazoles, in particular isoxaflutole, diketonitriles, in particular 2-cyano-3-cyclopropyl-1-(2-CH₃SO₂-4-CF₃ phenyl)propan-1,3-dione and 2-cyano-3-cyclopropyl-1-(2-CH₃SO₂-4,2,3-CI₂ phenyl)propan-1,3-dione, triketones, in particular sulcotrione or mesotrione, and pyrazolines.

11. Method according to Claim 10, characterized in that the concentration of HPPD inhibitors is between 0.5 mg/l and 50 mg/l, more preferably between 1 mg/l and [lacuna] mg/l.

12. Method for preparing transgenic plants comprising a heterologous gene integrated into their genome, which method comprises a method for transforming plant cells by introducing a heterologous gene into said plant cells with a gene for tolerance to HPPD inhibitors as a selection marker, said method comprising the steps of:

- 15 a) preparing and culturing competent plant cells capable of receiving the heterologous gene in a suitable medium,
 - b) transforming the competent cells with the heterologous gene and the selection marker,
 - c) growing and selecting the transformed cells comprising the heterologous
 - 20 gene in a suitable medium,
 - d) regenerating plants from the transformed cells selected in one or more suitable media and, where appropriate,
 - e) producing and recovering the seeds of the fertile transformed plants, then producing novel varieties of transgenic plants which have stably integrated the
 - 25 heterologous gene into their genome, in conventional selection programmes,
- characterized in that a step for bleaching the competent plant cells is carried out before the transformation step (b), by introducing a suitable amount of HPPD inhibitor into the suitable culture medium of the competent plant cells.

13. Method according to one of Claims 2 to 12, characterized in that the selection marker gene is eliminated by crossing the transformed plants comprising the heterologous gene and the selection marker gene with a nontransformed variety of the same plant.

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